A New Species of Dicyemid Mesozoan (Dicyemida: Dicyemidae) from *Sepioteuthis lessoniana* (Mollusca: Cephalopoda), with Notes on *Dicyema orientale*

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A new species of dicyemid mesozoan, *Dicyema koshidai* sp. nov., is described from Shiroika, *Sepioteuthis lessoniana* Lesson, 1830, collected off Uozu in Toyama Bay and off Mihonoseki in the Sea of Japan. The dicyemid is a large species that reaches a length of about 5000 µm. The vermiform stages are characterized as having 32–40 peripheral cells, a conical calotte, and an axial cell that extends to the middle of the metapolar cells. Infusoriform embryos consist of 37 cells; two nuclei are present in each urn cell and the refringent bodies are solid. The dicyemids live in folds of the renal appendages of the cephalopod host. This is the second dicyemid species to be described from *S. lessoniana*. Additional details of the first species, *D. orientale* Nouvel and Nakao, 1938, are described. The dicyemid fauna in *S. lessoniana* is discussed.
Key Words: cephalopods, dicyemids, infusoriform embryos, mesozoans, parasites, renal organs, *Sepioteuthis lessoniana*, vermiform embryos.

Introduction

The first record of dicyemids in Japan was published by Nouvel and Nakao (1938). They described *Dicyema misakiense* Nouvel and Nakao, 1938 from *Octopus vulgaris* Lamarck, 1798, and *D. orientale* Nouvel and Nakao, 1938 from *Sepioteuthis lessoniana* Lesson, 1830. Nouvel (1947) later described *D. acuticephalum* Nouvel, 1947 from *O. vulgaris* and identified a dicyemid species from *Sepia esculenta* Hoyle, 1885 as *Pseudicyema truncatum* Whitman, 1883, which had been described earlier in Europe. Subsequently, two dicyemid species, *D. japonicum* Furuya and Tsuneki, 1992 and *D. clavatum* Furuya and Koshida, 1992, were described from *O. vulgaris* and *Callistoctopus minor* (Sasaki, 1920), respectively (Furuya *et al.* 1992a). Recently, Furuya (1999) reported fourteen new species of dicyemid from six cephalopod species, *Amphioctopus fangsiao* d'Orbigny, 1840, *C. minor*, *Octopus hongkongensis* Hoyle, 1885, *Enteroctopus dofleini* (Wülker, 1910), *Sepia esculenta*, and *S. lycidas* Gray, 1849, caught off the coasts of Japan.

We examined the dicyemids that were found in the renal sacs of the bigfin reef squid, *Sepioteuthis lessoniana*. The species of squid inhabits inshore waters and is widely distributed throughout the Indo-West Pacific from the Indian Ocean to the western and central Pacific (Nesis 1982; Segawa 1987). We collected squid from six

localities in inshore waters of southwestern Japan and found two dicyemid species. One is *Dicyema orientale*. A second undescribed species was discovered in *S. lessoniana* collected off Uozu in Toyama Bay and off Mihonoseki, Shimane Prefecture, in the Sea of Japan. The present paper describes this new species and provides a revised, detailed description of *D. orientale*.

Materials and Methods

In this study 113 individuals of *Sepioteuthis lessoniana* were examined for dicyemids from 1989 to 2004. Host specimens were obtained from fish markets and fishermen. The size, sex, and locality of each squid are indicated in Appendix. A complex of three subspecies of *Sepioteuthis lessoniana* has been documented (Izuka *et al.* 1994, 1996). These have been named informally as Akaika, Shiroika, and Kuaika. The specimens examined in this study were identified as Shiroika based on their distributions and arrangement of chromatophores on the funnel.

When dicyemids were detected in the kidney of a host cephalopod, small pieces of renal appendages with attached dicyemids were removed and smeared on glass microscope slides. The smears were fixed immediately in Bouin's fluid for 24 hr and then stored in 70% ethyl alcohol. Most of them were stained in Ehrlich's hematoxylin and counterstained in eosin. Stained smears were mounted using Entellan (Merck). Dicyemids were observed with a light microscope (Olympus BH-2) at magnifications up to 2000×. Measurements and drawings were made with the aid of an ocular micrometer and a drawing tube (Olympus U-DA), respectively. The renal organs were also fixed with Bouin's solution. The fixed organs were embedded in paraffin and sectioned transversely. The sections were stained with hematoxylin-eosin and observed histologically.

Nouvel (1948), Short and Damian (1966), Furuya *et al.* (1992b), and Furuya (1999) give the terminology for cell names used in the description of infusoriform larvae.

Syntypes of the dicyemids are deposited in the Osaka University Museum, Toyonaka, Osaka, Japan (OUM), the Santa Barbara Museum of Natural History, Santa Barbara, California, USA (SBMNH), and in the auther's (HF) collection.

Taxonomy

Dicyema koshidai sp. nov.
(Figs 1–4, Table 1, Appendix)

Diagnosis. Large-sized dicyemids, body lengths typically not exceeding 5000 μm. Peripheral cell number of vermiform stages (i.e., vermiform embryo, nematogen, and rhombogen) 32–40, i.e., 4 propolars, 4 metapolars, and 24–32 trunk cells. Calotte relatively small, conical. Infusoriform embryos consisting of 37 cells; urn cells with 2 nuclei each.

Description. *Nematogens* (Figs 2b, 3a, c). Body slender; lengths ranging from 1000 to 5000 μm, widths from 50 to 70 μm. Peripheral cell number 32–40, i.e., 4 propolars, 4 metapolars, 2 parapolars, 20–28 diapolars, and 2 uropolars. Calotte bluntly
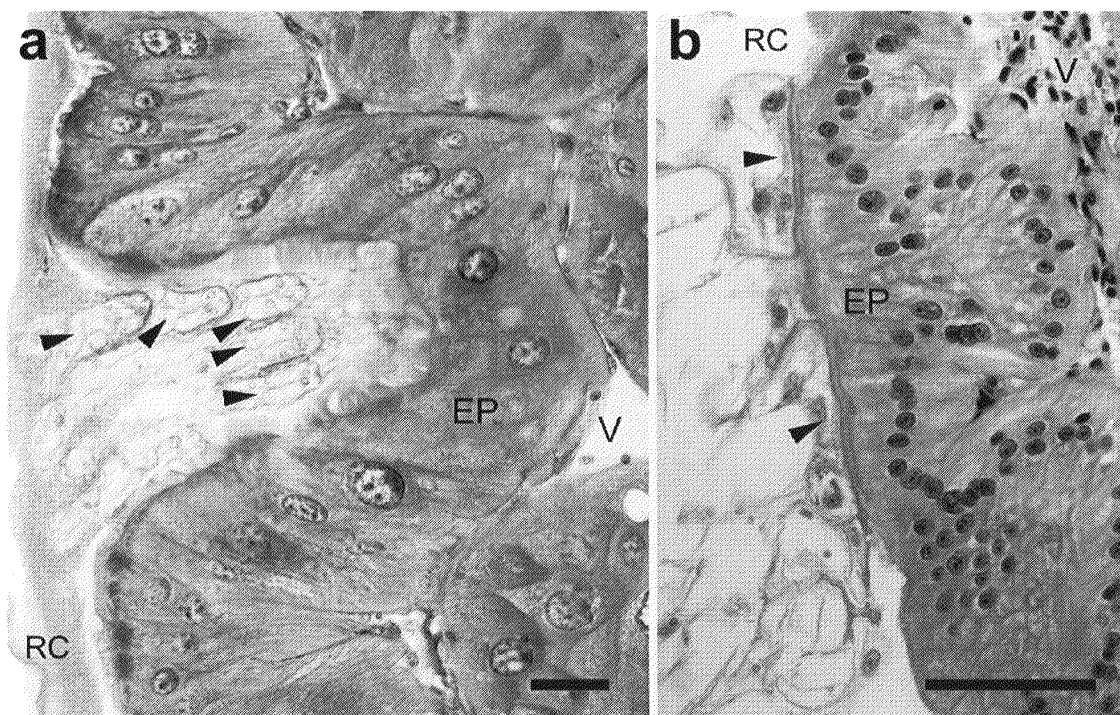
Fig. 1. Light micrographs of *Dicyema koshidai* sp. nov. (a) and *D. orientale* (b) in stained sections of renal organs of *Sepioteuthis lessoniana*. Note that the cone-shaped calotte and the anterior part of the body of *D. koshidai* are inserted into folds in the renal tissue (arrowheads in Fig. 1a). In contrast, *D. orientale*, with a slightly inflated, disc-shaped calotte, are attached to the surface of the renal appendages (arrowheads in Fig. 1b). Abbreviations: EP, epithelial cells of renal appendages; RC, renal coelom; V, vein. Scale bars: 50 μm.

rounded, conical. Cilia on calotte short, about 7 μm long, oriented forward. Cytoplasm of both propolar and metapolar cells more conspicuously stained with hematoxylin than trunk cells. Propolar cells and their nuclei smaller than metapolar cells and their nuclei, respectively. Trunk mostly uniform in width. Trunk cells arranged in opposed pairs. Axial cell cylindrical, tapered anteriorly, extending forward to middle of metapolar cells. In axial cell of large individuals 30–100 vermiform embryos present. Accessory nuclei few, sometimes seen in peripheral trunk cells.

Vermiform embryos (Figs 2c, 3f, g). Full-grown vermiform embryos medium-sized; lengths ranging from 80 to 130 μm, widths from 14 to 20 μm; peripheral cell number 32–40 (Table 1). Anterior end of calotte tapered slightly, bluntly rounded. Trunk cells arranged in opposed pairs. Axial cell tapered anteriorly, sometimes pointed, extending forward to base of metapolar cells, containing 4–8 agametes in full-grown embryos. Axial cell nucleus typically located in center of axial cell.

Rhombogens (Figs 2a, 3b, d, e). Slightly stockier than nematogens, otherwise generally similar in shape and body proportions; lengths ranging from 1000 to 5000 μm; widths from 60 to 80 μm. Peripheral cell number 32–39 (Table 1). Calotte conical as in nematogens. Shape and anterior extent of axial cell similar to those of nematogens. Number of infusorigens present in axial cell 4–13. In axial cells of large individuals, 100–300 infusoriform embryos typically present. Accessory nu-
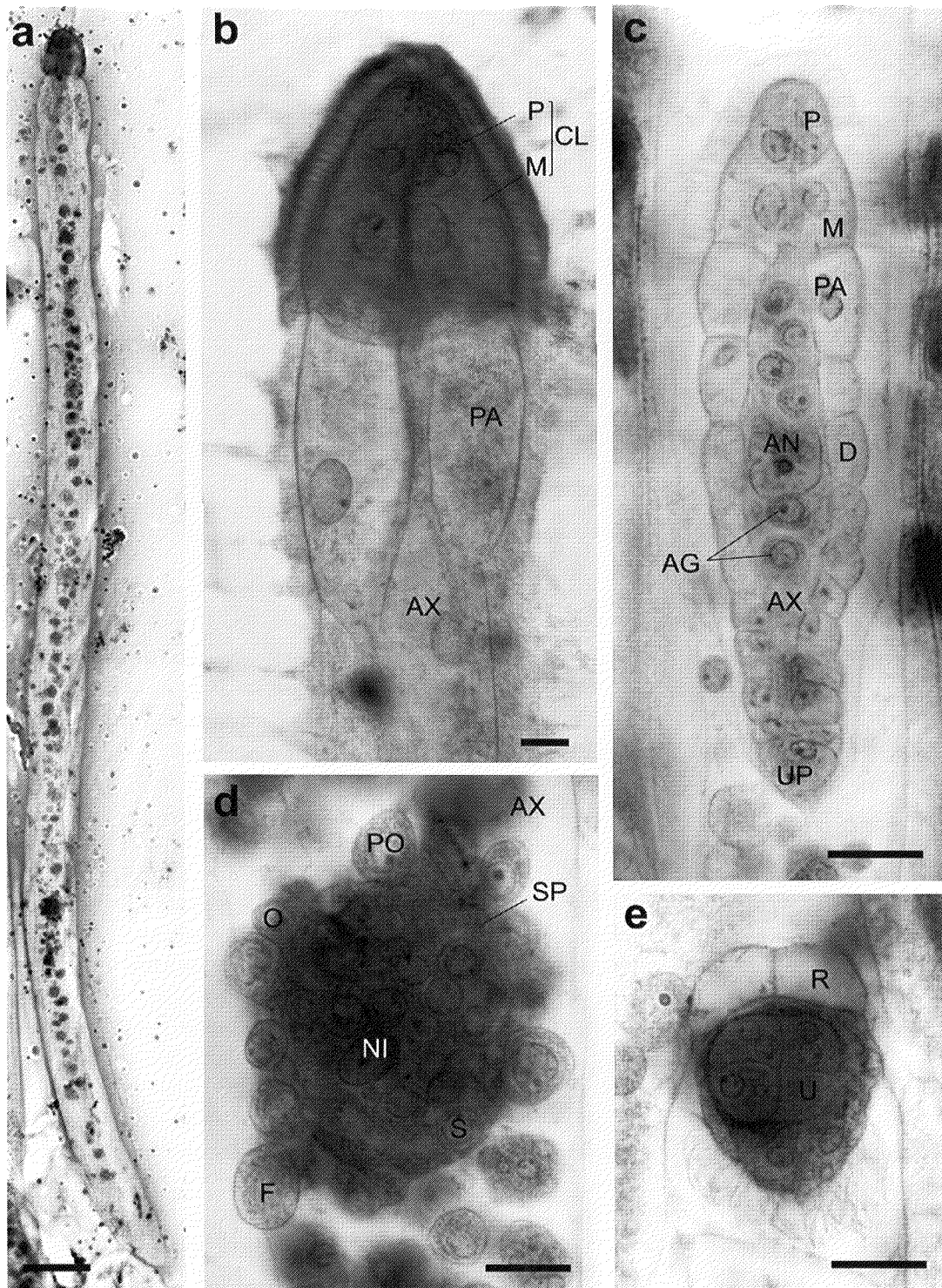
48 H. Furuya and K. Tsuneki

Fig. 2. *Dicyema koshidai* sp. nov., syntypes on slide OUM-ME-00001. a, Rhombogen, entire; b, nematogen, anterior region; c, vermiform embryos within axial cell; d, infusorigen; e, infusoriform embryo, horizontal section. Abbreviations: AG, agamete (axoblast); AN, axial cell nucleus; AX, axial cell; CL, calotte; D, diapolar cell; F, fertilized egg; M, metapolar cell; NI, nucleus of infusorigen; O, oogonium; P, propolar cell; PA, parapolar cell; PO, primary oocyte; R, refringent body; S, spermatogonium; SP, sperm; U, urn cell; UP, uropolar cell. Scale bars: 100 μm for a; 10 μm for b–e.

Table 1. Number of peripheral cells in *Dicyema koshidai* sp. nov.

Cell numberc	Number of individuals examined		
	Vermiform embryos	Nematogens	Rhombogens
32	5	3	4
33	2	2	3
34	6	5	5
35	4	3	3
36	5	5	6
37	8	6	4
38	15	10	9
39	6	3	1
40	1	1	0

clei occasionally present in peripheral cells.

Infusorigens (Fig. 2d). Large-sized. Axial cell usually irregular in shape. In mature infusorigens ($n=20$), number of external cells (oogonia and primary oocytes) 31–115 (mode, 49), number of internal cells (spermatogonia, primary spermatocytes, and secondary spermatocytes) 34–109 (mode, 58), and number of sperm 12–37 (mode, 21). Diameter of fertilized eggs 9.9 μm; diameter of sperm 2.4 μm.

Infusoriform embryos (Figs 2e, 4), Ovoid, rounded to bluntly pointed posteriorly. In full-grown embryos ($n=50$), length (excluding cilia) 26.4±1.7 μm (mean±S.D.), length-width-height ratio 1:0.82:0.71. Cilia at posterior end, 5.0 μm long. Refringent bodies present, solid, about same size as single urn cell, occupying anterior 40% of embryo length when viewed laterally (Fig. 4c). Cilia projecting from ventral internal cells into urn cavity (Fig. 4c). Cytoplasm of dorsal internal cells transparent. Capsule cells with many large, granular inclusions. Apical cell nuclei located in antero-ventral part of cell (Fig. 4b, c). Nuclei of second ventral cells small and pycnotic. Full-grown infusoriform embryos ($n=50$) consisting of 37 cells, i.e., 33 somatic and 4 germinal cells. Somatic cells composed of several varieties: external cells covering large part of anterior and lateral surfaces of embryo (2 enveloping cells); external cells with cilia on external surfaces (2 paired dorsal cells, 1 median dorsal cell, 2 dorsal caudal cells, 2 lateral caudal cells, 1 ventral caudal cell, 2 lateral cells, and 2 posteroventral lateral cells); external cells with refringent bodies (2 apical cells); external cells without cilia (2 first ventral cells, 2 second ventral cells, 2 third ventral cells, and 1 couvercle cell); internal cells with cilia (2 ventral internal cells); and internal cells without cilia (2 dorsal internal cells, 2 capsule cells, and 4 urn cells). Each of four urn cells containing 1 germinal cell plus 2 nuclei (Figs 1e, 4c). Nuclei of third ventral cells pycnotic. All somatic nuclei typically becoming pycnotic as infusoriform embryos mature.

Type series. Syntypes: OUM-ME-00001 (1 slide, OUM); SBMNH-353227 (1 slide, SBMNH); No. LE480 (5 slides) (author's (HF) collection). Type locality: Japan, Toyama Prefecture, Toyama Bay, Off Uozu, 36°41′N, 137°23′E. Host: *Sepioteuthis lessoniana* (Cephalopoda: Teuthoidae: Loliginidae), mature male, 15.8 cm ML (LE480). Site of infection: Within renal sacs; anterior ends (calottes) inserted into crypts of the renal appendages (Fig. 1).
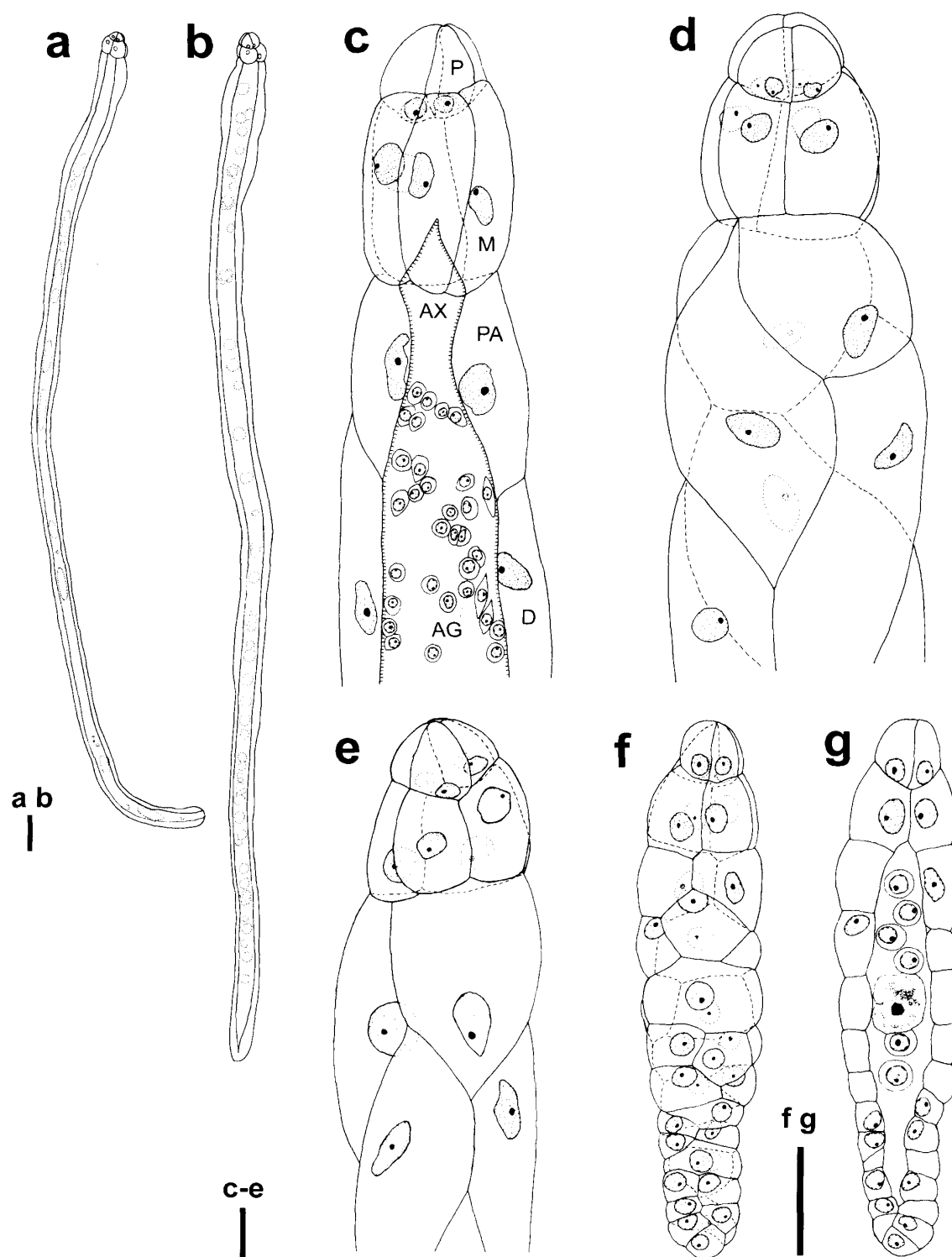
Fig. 3. *Dicyema koshidai* sp. nov., syntypes on slide OUM-ME-00001. a–b, Vermiform stages, entire—a, nematogen; b, rhombogen; c–e, vermiform stages, anterior region—c–d, nemato-gens; e, rhombogen; f–g, vermiform embryos within axial cell (cilia omitted). Abbreviations: AG, agamete (axoblast); AX, axial cell; D, diapolar cell; M, metapolar cell; P, propolar cell; PA, parapolar cell. Scale bars: 100 μm for a and b; 20 μm for c–e; 10 μm for f and g.
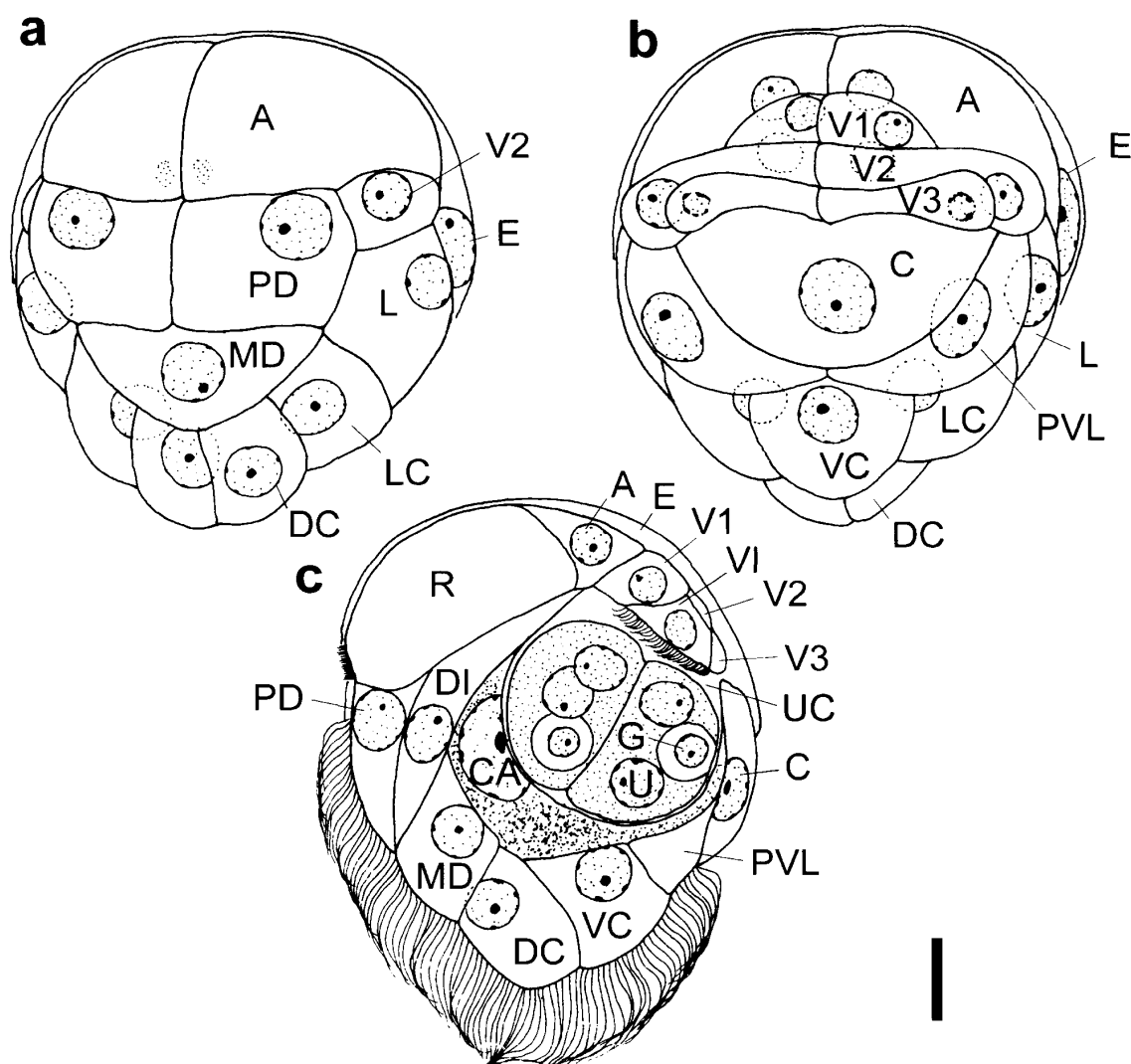
Fig. 4. *Dicyema koshidai* sp. nov., syntypes on slide OUM-ME-00001. a–c, Infusoriform embryos—a, dorsal view (cilia omitted); b, ventral view (cilia omitted); c, sagittal section. Abbreviations: A, apical cell; C, couvercle cell; CA, capsule cell; DC, dorsal caudal cell; DI, dorsal internal cell; E, enveloping cell; G, germinal cell; L, lateral cell; LC, lateral caudal cell; MD, median dorsal cell; PD, paired dorsal cell; PVL, posteroventral lateral cell; R, refringent body; U, urn cell; UC, urn cavity; VC, ventral caudal cell; VI, ventral internal cell; V1, first ventral cell; V2, second ventral cell; V3, third ventral cell. Scale bar: 5 μm.

Other material examined. A number of dicyemids found from 16 (including LE480) host cephalopods, *Sepioteuthis lessoniana*, obtained in Toyama Bay (Uozu) and Sea of Japan (Mihonoseki), with 14.2% in prevalence among 113 cephalopods examined in total (see Appendix).

Distribution. Toyama Bay and the coast of the Shimane Peninsula in the Sea of Japan.

Etymology. The species is named in honor of the late Dr. Yutaka Koshida (Professor Emeritus, Osaka University), who was an eminent zoologist and studied various invertebrates including dicyemids.

Remarks. *Dicyema koshidai* sp. nov. is characterized by a conical calotte, a large number of peripheral cells, and a variable number of peripheral cells that ranges from 32 to 40 with a mode of 38 (Table 1). In these respects *D. koshidai* is easily distinguished from *D. orientale*.

In the genus *Dicyema*, both *D. erythrum* Furuya, 1999 and *D. australis* Penchaszadeh, 1968 have more than 32 peripheral cells. The body of *D. erythrum* consists of 26–36 (typically 32–34) peripheral cells. *Dicyema erythrum* is very similar to *D. koshidai* in the shape of the calotte in vermiform stages and in both cell number and cellular composition of infusoriform embryos; however, *D. erythrum* can be distinguished from *D. koshidai* based on the typical number of peripheral cells and the typical number of agametes (3) in full-grown vermiform embryos (Furuya 1999). In addition, axial cells of *D. erythrum* extend forward to the base of the propolar cells, whereas those of *D. koshidai* end forward to the middle of metapolar cells.

The body of *D. australis* consists of 37–40 (typically 39) peripheral cells. Thus, *D. australis* differs from *D. koshidai* in the typical number of peripheral cells (Penchaszadeh 1968). In addition, *D. australis* differs in having less than five agametes in full-grown vermiform embryos and a single nucleus in the urn cells of infusoriform embryos.

The size and number of infusorigens are characteristics of dicyemid species (Furuya *et al.* 1993). There is a negative curvilinear relationship between the number of infusorigens per rhombogen and the number of gametes per infusorigen (Furuya *et al.* 2003b). Two distinct groups of dicyemid species are apparent. One type forms a small number of infusorigens and produces a relatively large number of gametes (four to 70) per infusorigen. The other type tends to produce a large number of infusorigens, each of which has at most 20 gametes per infusorigen, as in *D. orientale*. Rhombogens of *D. koshidai* have a relatively large number of large-sized infusorigens, and thus they do not belong to either of these types. There are, in fact, a few exceptional species in which the rhombogens produce large numbers of infusorigens and each infusorigen has a large number of gametes (Furuya *et al.* 2003b). Infusorigens of *D. koshidai* belong to this rare third type.

With regard to cellular composition and cell number the infusoriform embryos of *D. koshidai* are of the typical type (Furuya, Hochberg *et al.* 2004). However, its embryos are exceptional in that the nucleus of the apical cell is located in the antero-ventral part of the cell.

In the present host species, geographical variation in morphology or cell number of *D. koshidai* was not observed in either vermiform stages or infusoriform embryos.

Dicyema orientale Nouvel and Nakao, 1938
(Figs 1, 5–7, Table 2, Appendix)

Diagnosis. Large-sized dicyemids, body length typically not exceeding 5000 μm. Peripheral cell number of vermiform stages (i.e., vermiform embryo, nematogen, and rhombogen) 22, i.e., 4 propolars, 4 metapolars, 2 parapolars, and 12 trunk cells. Calotte disc-shaped; cephalic swelling formed together with parapolar cells. Infusoriform embryos consisting of 37 cells; urn cells with 2 nuclei each.

Description. *Nematogens* (Figs 5a, 6c). Body hammer-like; lengths ranging
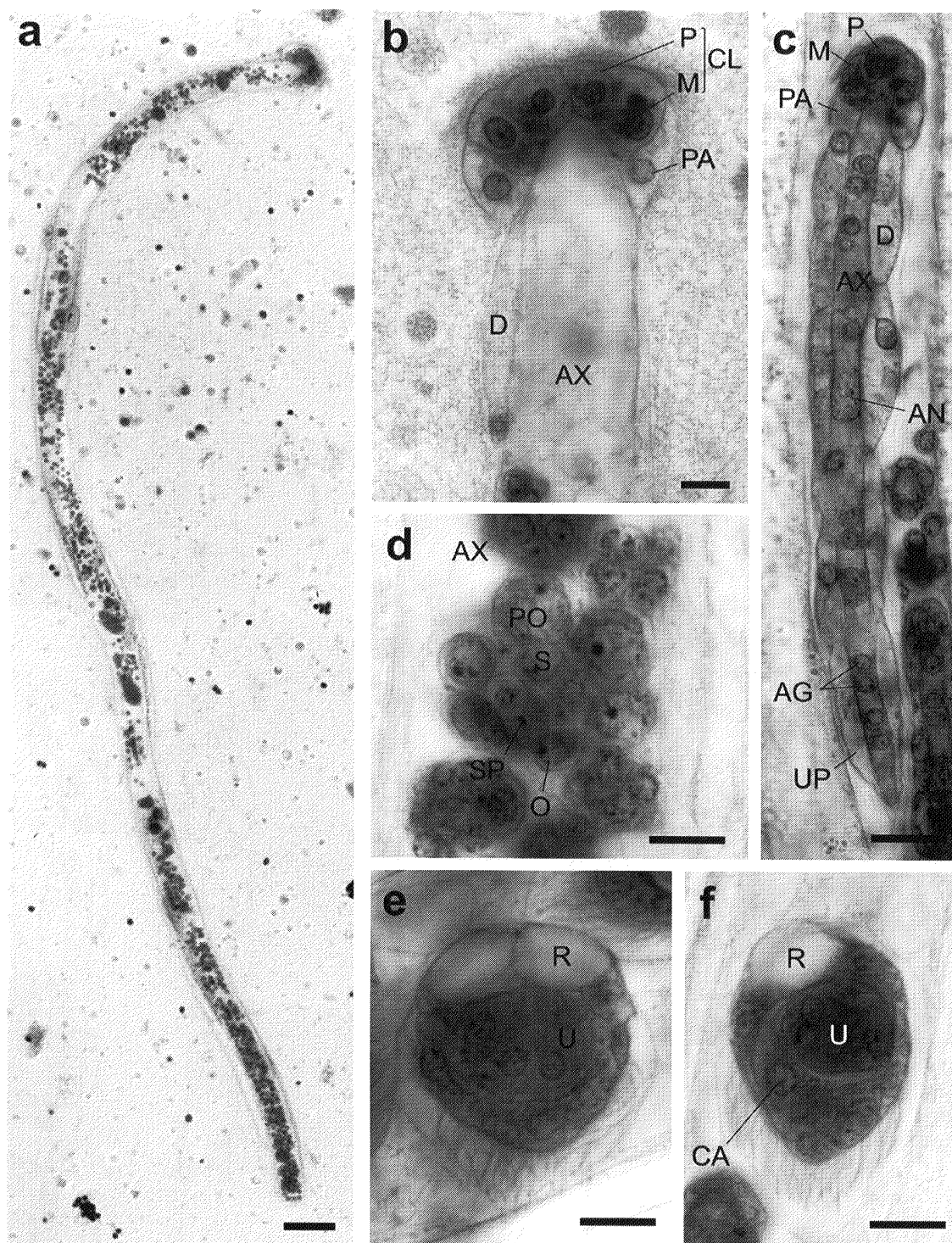
Fig. 5. *Dicyema orientale*, specimens on slide OUM-ME-00002. a, Nematogen, entire; b, rhombogen, anterior region; c, vermiform embryo within axial cell; d, infusorigen; e, f, infusoriform embryos—e, horizontal section; f, sagittal section. Abbreviations: AG, agamete (axoblast); AN, axial cell nucleus; AX, axial cell; CA, capsule cell; CL, calotte; D, diapolar cell; M, metapolar cell; O, oogonium; P, propolar cell; PA, parapolar cell; PO, primary oocyte; R, refringent body; S, spermatogonium; SP, sperm; U, urn cell; UP, uropolar cells. Scale bars: 100 μm for a; 10 μm for b–f.
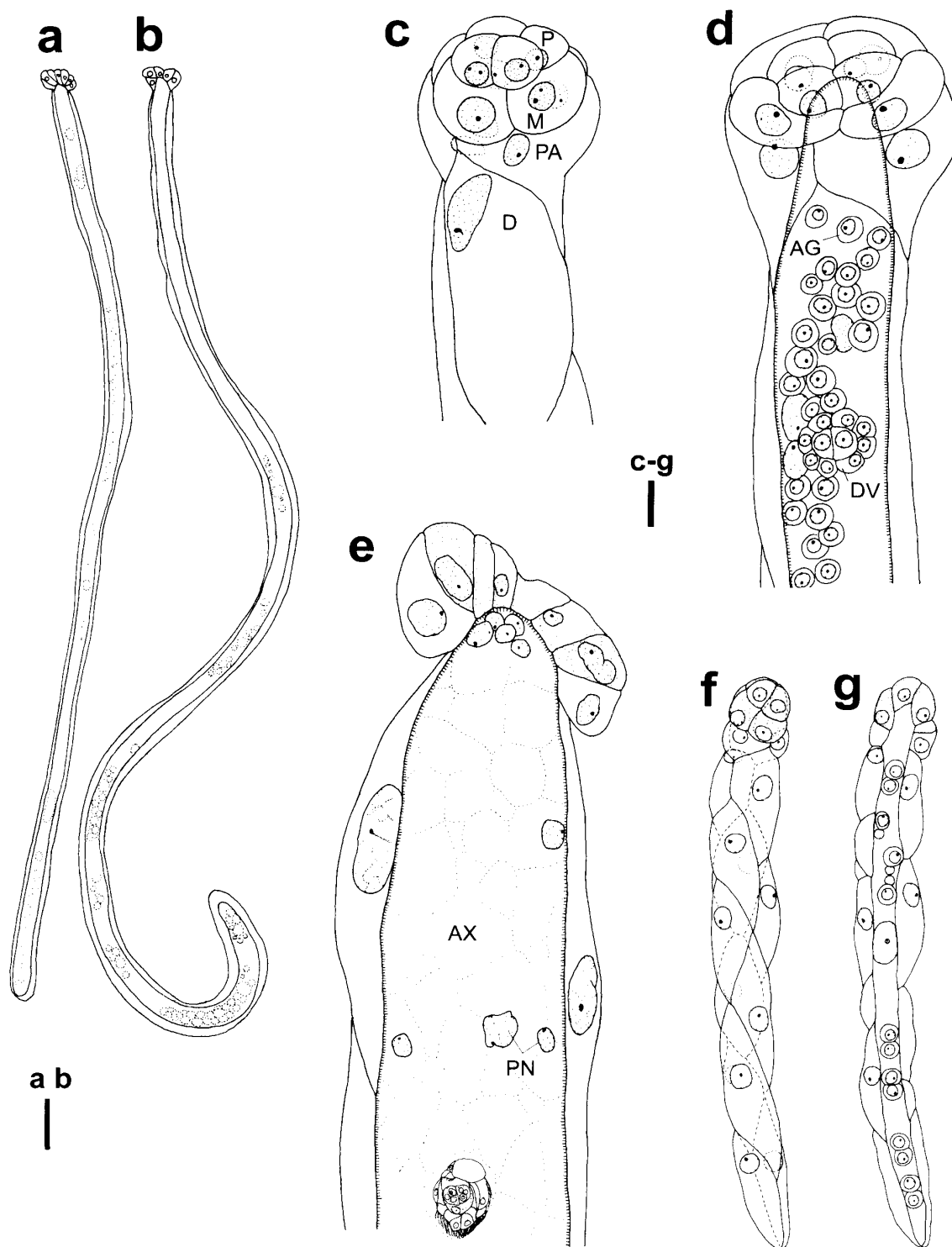
Fig. 6. *Dicyema orientale*, specimens on slide OUM-ME-00002. a–b, Vermiform stages, entire—a, nematogen; b, rhombogen; c–e, vermiform stages, anterior region—c–d, nematogens; e, rhombogen; f–g, vermiform embryos within axial cell (cilia omitted). Abbreviations: AG, agamete (axoblast); AX, axial cell; D, diapolar cell; DV, developing vermiform embryo; M, metapolar cell; P, propolar cell; PN paranucleus; PA, parapolar cell. Scale bars: 100 µm for a and b; 10 µm for c–g.

from 1000 to 5000 μm; widths from 60 to 80 μm. Peripheral cell number mostly 22, i.e., 4 propolars, 4 metapolars, 2 parapolars, 10 diapolars, and 2 uropolars. Calotte disc-shaped in large individuals. Cilia on calotte about 6 μm long, oriented forward. Cytoplasm of propolar and metapolar cells more conspicuously stained with hematoxylin than trunk cells. Propolar cells and their nuclei smaller than metapolar cells and their nuclei, respectively. Axial cell cylindrical, rounded anteriorly, extending forward to base of propolar cells. About 40 vermiform embryos typically present in axial cell of large individuals.

Vermiform embryos (Figs 5c, 6f, g). Full-grown vermiform embryos medium-sized; lengths ranging from 80 to 150 μm, widths from 12 to 15 μm; peripheral cell number 22 (Table 2). Anterior end of calotte rounded. Trunk cells arranged in opposed pairs. Axial cell rounded anteriorly, extending forward to base of propolar cells, as in nematogens, typically containing 8–13 agametes in full-grown embryos. Axial cell nucleus usually located in center or occasionally in anterior half of axial cell.

Rhombogens (Figs 5b, 6b, e). Slightly stockier than nematogens, otherwise generally similar to them in shape and body proportions; lengths ranging from 1000 to 5000 μm; widths from 60 to 100 μm. Peripheral cell number usually 22, occasionally 20 (Table 2). Cephalic enlargement composed of calotte and parapolar cells as in nematogens. Calotte disc-shaped in large individuals. Shape and anterior extent of axial cell similar to those of nematogens. Number of infusorigens present in axial cell of rhombogen ranging from 7 to 31. In axial cells of large individuals, 100–300 infusoriform embryos typically present. Uropolar cells verruciform. Accessory nuclei occasionally observed in trunk peripheral cells.

Infusorigens (Fig. 5d). Medium-sized. Axial cell of infusorigens usually rounded, diameter 12–18 μm. In mature infusorigens (n=50), number of external cells (oogonia and primary oocytes) 8–13 (mode, 10), number of internal cells (spermatogonia, primary and secondary spermatocytes) 2–5 (mode, 3), and number of sperm 6–17 (mode, 12). Diameter of fertilized eggs 11.8 μm; diameter of sperm 2.8 μm.

Infusoriform embryos (Figs 5e, f, 7a–c). Ovoid, rounded to bluntly pointed posteriorly. In full-grown embryos (n=100), length (excluding cilia) 25.7±1.9 μm (mean±S.D.); length-width-height ratio 1:0.84:0.80. Cilia at posterior end 8 μm long. Refringent bodies present, solid, relatively small, about same size as single urn cell, occupying about 25–30% of embryo length when viewed laterally (Fig. 5f). Cilia projecting from ventral internal cells into urn cavity (Fig. 7c). Capsule cells containing many large granules. Full-grown infusoriform embryos (n=100) consisting of 37 cells, i.e., 33 somatic and 4 germinal cells. Somatic cells composed of

Table 2. Number of peripheral cells in *Dicyema orientale*.

Cell number	Number of individuals examined		
	Vermiform embryos	nematogens	rhombogens
20	0	2	3
21	1	1	0
22	49	58	20

several varieties: external cells covering large part of anterior and lateral surfaces of embryo (2 enveloping cells); external cells with cilia on external surfaces (2 paired dorsal cells, 1 median dorsal cell, 2 dorsal caudal cells, 2 lateral caudal cells, 1 ventral caudal cell, 2 lateral cells, and 2 posteroventral lateral cells); external cells with refringent bodies (2 apical cells); external cells without cilia (2 first ventral cells, 2 second ventral cells, 2 third ventral cells, and 1 couvercle cell); internal cells with cilia (2 ventral internal cells); and internal cells without cilia (2 dorsal internal cells, 2 capsule cells, and 4 urn cells). Each urn cell containing 1 germinal cell plus 1 nucleus (Figs 5e, 7c). Nuclei of first ventral cells pycnotic. All somatic
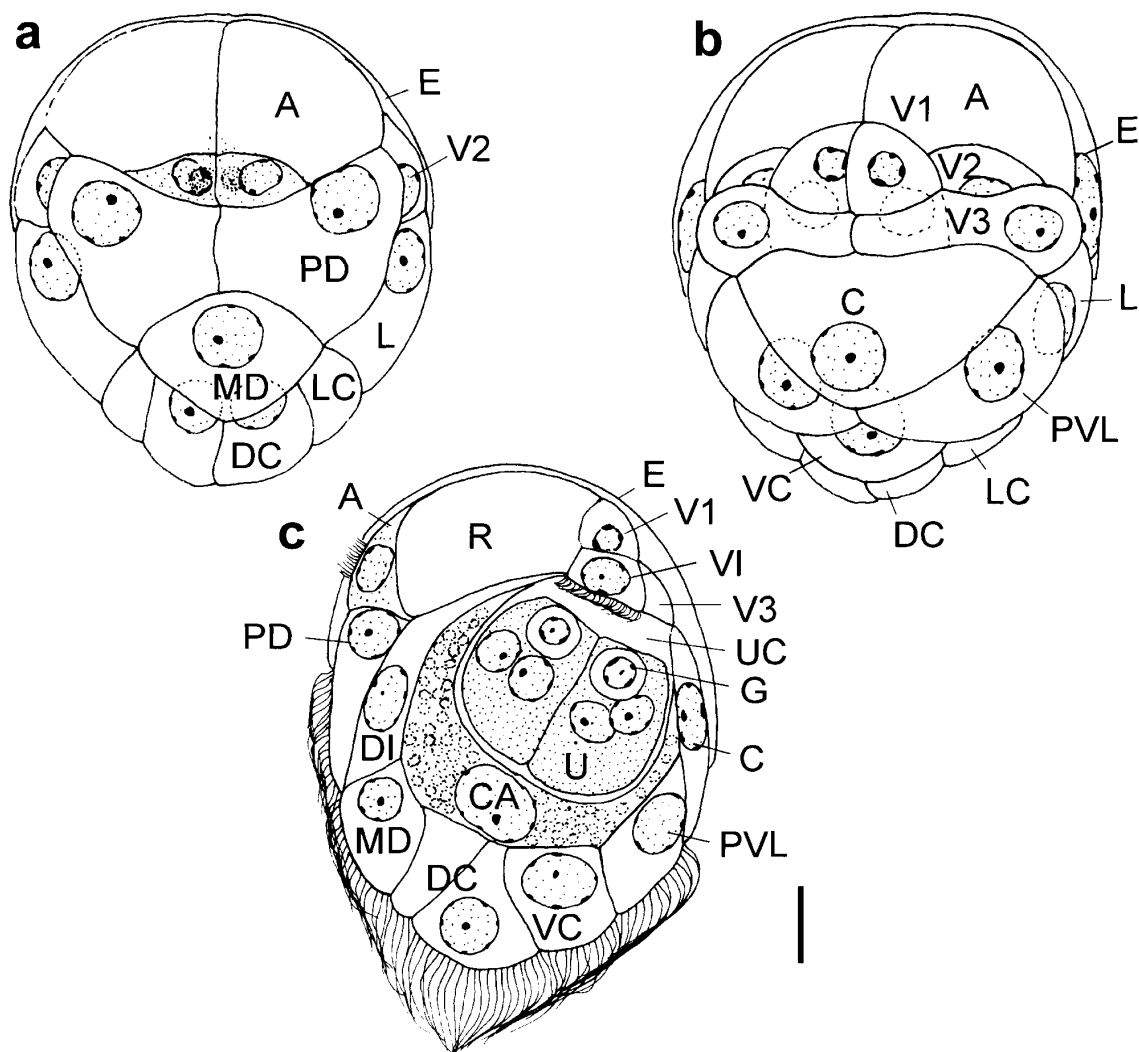

Fig. 7. *Dicyema orientale*, specimens on slide OUM-ME-00002. a–c, Infusoriform embryos—a, dorsal view (cilia omitted); b, ventral view (cilia omitted); c, sagittal section. Abbreviations: A, apical cell; C, couvercle cell; CA, capsule cell; DC, dorsal caudal cell; DI, dorsal internal cell; E, enveloping cell; G, germinal cell; L, lateral cell; LC, lateral caudal cell; MD, median dorsal cell; PD, paired dorsal cell; PVL, posteroventral lateral cell; R, refringent body; U, urn cell; UC, urn cavity; VC, ventral caudal cell; VI, ventral internal cell; V1, first ventral cell; V2, second ventral cell; V3, third ventral cell. Scale bar: 5 μm.

nuclei becoming pycnotic as infusoriform embryos mature.

Material examined. A number of dicyemids found from 55 host cephalopods, *Sepioteuthis lessoniana*, obtained in the Sea of Japan (Sakaiminato and Mihonoseki), the Inland Sea (Akashi), and the Pacific Ocean (Minabe), with 48.7% in prevalence among 113 cephalopods examined in total (see Appendix). Host: *Sepioteuthis lessoniana* (Cephalopoda: Teuthoidae: Loliginidae). Site of infection: Within renal sacs; attached to the surface of the renal appendages (Fig. 1). Of these material, several slides of dicyemids are deposited as voucher specimens in the following collections: OUM-ME-00002 (1 slide, OUM); SBMNH 353228 (1 slide, SBMNH); Nos. LE47 (5 slides), LE122 (5 slides), LE124 (5 slides), LE992 (1 slide), and LE1017 (2 slides) (author's (HF) collection). Description of *Dicyema orientale* given above is based on individuals removed from a mature male cephalopod of 38.5 cm ML (LE124) collected in Kii Strait off Minabe, Wakayama Prefecture.

Distribution. Western Honshu, Japan.

Discussion

Pelagic cephalopods rarely harbor dicyemids. A few dicyemid species have been reported from two neritic squid species, namely, *Sepioteuthis lessoniana* in Japanese waters (Nouvel and Nakao 1938) and *Loligo* sp. in the Indian Ocean (Kalavati and Narasimhamurti 1980), and from the oceanic squid *Todarodes pacificus* Steenstrup, 1880 in Toyama Bay (Furuya and Tsuneki 2003). Bayard H. McConnaughey found dicyemids in *Loligo opalescens* off California but did not describe the species (F. G. Hochberg pers. comm.). Dicyemids produce a dispersal larva, the infusoriform larva, that is not planktonic. Infusoriform larvae actively swim close to the bottom for only a few days *in vitro* (McConnaughey 1951; Furuya unpublished data). In the anterior region of an embryo there is a pair of unique cells called apical cells, each containing a refringent body composed of a hydrated magnesium salt of inositol hexaphosphate (Lapan 1975). Its high specific gravity imparts a negative buoyancy to the dispersal larvae. Some species have no refringent bodies or only mucoid ones (Short 1971; Furuya 1999). McConnaughey (1951) and Lapan (1975) suggested that the role of the refringent bodies is to help the larvae remain near the sea bottom, where they can encounter another host. Most of the cephalopod hosts that harbor dicyemids are indeed benthic species. Although the life history of squid hosts harboring dicyemids is not fully clarified, a benthonic stage may be involved.

In this study, two dicyemid species, *Dicyema orientale* and *D. koshidai*, were found in 71 out of 113 individuals of *Sepioteuthis lessoniana*. Prevalence of dicyemids in *S. lessoniana* was 62.2%, which is relatively low. *Dicyema koshidai* has been found from hosts obtained in Toyama Bay and along the shores of the Shimane Peninsula, but never off the Pacific coast of Japan. *Dicyema orientale* was described from *S. lessoniana* collected in waters close to the Misaki Marine Biological Station of the University of Tokyo, which is located on the southeastern coast of Honshu (Nouvel and Nakao 1938). In contrast to *D. koshidai*, *D. orientale* has been found in hosts obtained from the Sea of Japan, the Inland Sea, and the Pacific Ocean. In Japan this species is the dominant dicyemid in *S. lessoniana* found along the western coast of Honshu Island.

Off the coast of the Shimane Peninsula two dicyemid species have been found in *S. lessoniana*; however, they have not been found to co-occur in the renal sac of a single host individual. In cephalopods harboring more than one dicyemid species, generally two or three dicyemid species coexist in the renal sac. Dicyemids with similar calotte shapes are only rarely found in the same host cephalopod species, such as *Octopus joubini* and *O. vulgaris* (Furuya *et al.* 2003a). Similar types of dicyemid rarely, if ever, share the same renal sac, because the dominant or more common species occupies the niche, competitively excluding the other species. When more than one dicyemid species is present in a single host, the calotte shapes are usually different from each other. *Dicyema orientale* and *D. koshidai* indeed have their own niches (on the surface vs. in the folds) that are determined by differences in calotte shape (disc-shaped vs. conical).

Izuka *et al.* (1994, 1996) suggested that *S. lessoniana* forms a complex that consists of three species or subspecies, Akaika, Kuaika, and Shiroika. In this study all squid specimens belonged to Shiroika. Dicyemids have a narrow host-specificity (Furuya 1999). As far as dicyemid species are concerned, there might be two different populations in Shiroika.

No dicyemids were found in 42 examined individuals of *S. lessoniana*. Smaller or younger cephalopods of a host species generally do not harbor dicyemids (Furuya *et al.* 1992a). There is a direct relationship between host size and dicyemid occurrence. Depending on the species of host cephalopod there is probably a specific size at which the species is infected with dicyemids. For instance, dicyemids are not detected in small individuals of the medium-sized octopus, *Octopus vulgaris*, which is similar in size to the small-sized octopus, *A. fangsiao*, which harbors dicyemids. In *S. lessoniana*, however, the relationship between the host size and dicyemid presence differs based on geographical location (Table 3). At Minabe (Pacific coast) and Akashi (Inland Sea), dicyemids are rarely, if ever, found in small host individuals. At Uozu, Sakaiminato, and Mihonoseki (Sea of Japan), the incidence of dicyemids is high even in small host individuals.

Even in large host individuals of *S. lessoniana*, no dicyemids have been de-

Table 3. Prevalence of dicyemids in each size of the host cephalopod *Sepioteuthis lessoniana* collected off Japan.

Localities	Mantle length (cm)									
	<10	<15	<20	<25	<30	<35	<40	<45	<50	<55
Minabe			0% (3)	50.0% (12)	94.7% (19)	77.7% (9)	81.3% (16)	100.0% (4)	50.0% (2)	100% (1)
Akashi		0% (2)	0% (2)	0% (4)	100% (2)					
Uozu		100% (8)	100% (5)							
Sakaiminato and Mihonoseki	0% (4)	62.5% (8)	50% (2)							
Okinawa				0% (4)	0% (4)	0% (2)				

The number of hosts is given in parentheses.

tected in Okinawa. This suggests that no dicyemids infect *S. lessoniana* in this area. Okinawa is the center of the subtropical Ryukyu Islands chain. Hochberg (1990) proposed that dicyemids are restricted to temperate and sub-boreal waters. The degree of infection of host individuals decreases with decreasing latitude from temperate regions, where individuals of cephalopod species are 100% infected, to tropical regions, where no records of infected cephalopods have been traced. His proposal is based on negative findings for dicyemids in cephalopods from Hawaii and the Marshall Islands (McConnaughey 1949). Dicyemids also have not been found in some Japanese subtropical octopus species (Furuya, Ota *et al.* 2004). On the contrary, Finn (1997) studied the presence or absence of dicyemids in several octopus species distributed in tropical to cold waters in Australia and his results do not support Hochberg's (1990) proposal. In Australia tropical octopuses, such as *Callistoctopus alpheus* (Norman, 1993), *C. dierythraeus* (Norman, 1993), and *C. ornatus* (Gould, 1852), harbor dicyemids, although *O. ornatus* does not harbor dicyemids in Japanese waters.

Dicyemids have not been detected in either *Octopus cyaneus* Gray, 1849 or *Callistoctopus ornatus*, which live on coral reefs in subtropical seas (Okinawa) (Furuya, Ota *et al.*, 2004). Similarly, no dicyemids are found in an undescribed *Amphioctopus* from Hawaii (F. G. Hochberg pers. comm.). These octopuses use a hole or gap in a rock or coral as a living space. Dicyemids may not infect cephalopods that live in corals and rocks even though they are benthonic in habitat. The largest cuttlefish, *Sepia latimanus* Quoy and Gaimard, 1832, harbors dicyemids (Furuya and Tsuneki 2003; Finn 1997). This cuttlefish inhabits coral reefs in subtropical seas, often swimming over the coral but usually lying on the sand bottom. If *Sepioteuthis lessoniana* are infected with dicyemids when in contact with the bottom, their renal organs may be available for dicyemids as a habitat. The fact that dicyemids are not found in *S. lessoniana* off Okinawa is possibly attributed to the fact that benthonic stage of this squid is associated with coral habitats and does not occur on sandy substrates.

Acknowledgments

Appendix: Dicyemids from Cephalopod Hosts

In the following list, the dicyemids found in the present study from the host cephalopod, *Sepioteuthis lessoniana*, are arranged under respective localities fol-

60   H. Furuya and K. Tsuneki

lowed by the cephalopod data, i.e., individual code, dorsal mantle length (cm), sex, and date of examination.

Minabe (Kii Strait, Pacific coast)
Dicyema orientale

LE47, 27.5, ♂, 20.12.1989	LE122, 29.5, ♀, 15.05.1992	LE123, 23.5, ♀, 15.05.1992
LE124, 38.5, ♂, 15.05.1992	LE241, 29.4, ♂, 13.12.1994	LE242, 25.6, ♂, 13.12.1994
LE390, 34.5, ♂, 04.03.1997	LE391, 29.8, ♂, 04.03.1997	LE392, 28.4, ♂, 04.03.1997
LE393, 27.0, ♂, 04.03.1997	LE394, 36.0, ♂, 04.03.1997	LE395, 32.8, ♂, 04.03.1997
LE396, 32.5, ♂, 04.03.1997	LE397, 32.3, ♂, 04.10.1997	LE398, 29.5, ♂, 04.10.1997
LE399, 27.3, ♀, 04.10.1997	LE400, 22.5, ♀, 04.10.1997	LE401, 25.6, ♂, 04.10.1997
LE404, 46.0, ♂, 28.04.1997	LE406, 36.8, ♂, 28.04.1997	LE408, 22.6, ♂, 28.04.1997
LE471, 37.0, ♂, 15.05.1998	LE472, 41.0, ♂, 15.05.1998	LE473, 35.0, ♂, 15.05.1998
LE475, 43.0, ♂, 15.05.1998	LE476, 28.0, ♂, 20.05.1998	LE477, 32.0, ♂, 20.05.1998
LE464, 41.5, ♂, 24.05.1999	LE465, 43.0, ♂, 24.05.1999	LE466, 50.8, ♂, 24.05.1999
LE468, 35.0, ♂, 18.01.1999	LE469, 34.0, ♂, 18.01.199	LE553, 34.2, ♂, 17.04.2000
LE554, 35.0, ♂, 17.04.2000	LE556, 38.2, ♂, 17.04.2000	LE815, 28.0, ♀, 29.06.2000
LE823, 28.4, ♂, 10.10.2002	LE824, 26.8, ♂, 10.10.2002	LE890, 27.8, ♂, 11.06.2003
LE891, 26.5, ♀, 11.06.2003	LE893, 20.5, ♂, 11.06.2003	LE953, 22.0, ♀, 23.10.2003
LE954, 25.2, ♂, 23.10.2003	LE955, 20.3, ♀, 23.10.2003	LE956, 25.0, ♂, 23.10.2003
LE1013, 35.8, ♂, 29.01.2004	LE1014, 35.5, ♂, 29.01.2004	LE1015, 36.0, ♀, 29.01.2004
LE1016, 39.0, ♀, 29.01.2004	LE1017, 38.2, ♂, 29.01.2004.	

No dicyemid

LE405, 38.4, ♂, 28.04.1997	LE407, 29.4, ♂, 28.04.1997	LE463, 45.0, ♂, 24.05.1999
LE467, 21.8, ♂, 24.05.1999	LE470, 31.0, ♂, 18.01.1999	LE474, 38.0, ♂, 15.05.1998
LE552, 31.7, ♂, 17.04.2000	LE555, 35.4, ♂, 17.04.2000	LE816, 22.2, ♀, 29.06.2000
LE817, 21.5, ♀, 29.06.2000	LE818, 18.5, ♂, 29.06.2000	LE819, 17.5, ♀, 29.06.2000
LE820, 20.5, ♂, 10.10.2002	LE821, 21.2, ♂, 10.10.2002	LE822, 22.5, ♂, 10.10.2002
LE892, 19.7, ♀, 11.06.2003		

Akashi (the Inland Sea)
Dicyema orientale

LE1062, 29.5, ♂, 18.06.2004	LE1063, 29.0, ♂, 18.06.2004

No dicyemid

LE221, 20.8, ♂, 21.11.1994	LE240, 23.4, ♂, 08.12.1994	LE928, 10.5, ♀, 21.10.2003
LE929, 11.5, ♀, 21.10.2003	LE930, 17.0, ♂, 21.10.2003	LE931, 18.0, ♂, 21.10.2003
LE1009, 22.6, ♂, 26.11.2003	LE1010, 22.7, ♂, 26.11.2003	

Uozu (Toyama Bay)
Dicyema koshidai

LE478, 10.6, ♂, 26.01.2000	LE479, 17.5, ♂, 26.01.2000	LE480, 15.8, ♂, 26.01.2000
LE999, 16.0, ♂, 19.11.2003	LE1000, 15.0, ♀, 19.11.2003	LE1001, 14.5, ♂, 19.11.2003
LE1002, 14.2, ♀, 19.11.2003	LE1003, 15.2, ♀, 19.11.2003	LE1004, 14.5, ♂, 19.11.2003
LE1005, 14.6, ♂, 19.11.2003	LE1006, 14.5, ♂, 19.11.2003	LE1007, 13.2, ♀, 19.11.2003
LE1008, 13.2, ♂, 19.11.2003		

Sakaiminato (Shimane Peninsula, Sea of Japan)
Dicyema orientale

LE961, 14.2, ♀, 12.11.2003

No dicyemid

LE958, 12.3, ♀, 12.11.2003	LE959, 13.6, ♀, 12.11.2003	LE960, 14.0, ♀, 12.11.2003

Mihonoseki (Shimane Peninsula, Sea of Japan)
Dicyema koshidai
LE991, 13.4, ♂, 13.11.2003 LE993, 10.9, ♀, 13.11.2003 LE994, 11.2, ♀, 13.11.2003
Dicyema orientale
LE989, 17.3, ♀, 13.11.2003 LE992, 11.4, ♂, 13.11.2003
No dicyemid
LE990, 17.2, ♀, 13.11.2003 LE995, 9.7, ♀, 13.11.2003 LE996, 8.3, ♂, 13.11.2003
LE997, 9.2, ♀, 13.11.2003 LE998, 8.6, ♂, 13.11.2003
Okinawa (Ryukyu Islands)
No dicyemid
LE541, 31.2, ♀, 05.04.2000 LE542, 37.0, ♂, 05.04.2000 LE900, 27.8, ♂, 17.11.2003
LE901, 23.5, ♂, 17.11.2003 LE910, 20.8, ♂, 18.11.2003 LE911, 21.8, ♂, 18.11.2003
LE912, 29.0, ♂, 18.11.2003 LE913, 26.2, ♂, 18.11.2003 LE1011, 28.4, ♂, 08.01.2004
LE1012, 20.1, ♂, 08.01.2004

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